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13. ABSTRACT (Maximum 200 words)

To identify soldiers with a possible health risk when deployed rapidly to moderate altitude, oculometrics were investigated as an objective index of the hypoxic effect on the central nervous system. We previously described the effects of a 4-h exposure to real and simulated 4,300 m altitude on the pupillary light reflex as measured by the initial pupil diameter (PD), constriction amplitude (CA), and constriction latency (CL). Saccadic Velocity (SV) was measured as an index of extraocular motor function. Within 1 h of exposure PD was reduced 5% and CL 2%. After 24 h, the reductions progressed to 12% and 5%, respectively. These effects were due to hypoxia per se, readily reversible, not gender related, and independent of eye dominance. SV was not acutely affected. To determine whether oculomotor reflexes could serve as an acclimatization variable and an index of AMS severity, data were collected during the long-term phase of the field study conducted at Pikes Peak, CO (4,300 m). After sea-level, baseline (SLB) measurements were taken, 18 men (age range 19-33 yrs) were transported to Pikes Peak where they remained for 14 days. The same oculometric variables were measured on days 1-4, 6, 7, 9, 10, and 12 in addition to several classical measures of altitude acclimatization: environmental symptoms questionnaire (ESQ), heart rate (HR), pulse oximetry (SpO2), end-tidal PO2 and PCO2, and 24-h urinary epinephrine and norepinephrine concentrations. PD and CL decreased from SLB for days 1-4 and returned toward SLB; these changes paralleled changes in ventilatory and circulatory variables. CA decreased on days 1 and 2 and remained decreased for 12 days. SV increased over the first 6 days then returned toward SLB with continued exposure, similar to the changes in urinary catecholamines. With acclimatization, CL correlated with HR and SpO2; SV correlated with PCO2, HR, and SpO2. AMS severity peaked during days 2-4, returned toward SLB over the next 10 days, and correlated only with CL ($p=0.045$). Thus, it appears that selected pupillary and oculomotor measures can be used as additional indices of altitude acclimatization, even though there was no strong correlation of any oculomotor measurement with the severity of AMS.

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ALTITUDE (4300 m)**

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BACKGROUND

This report contains data collected during the last year of a three-year VA/DoD project to study the effects of energy deficit on work performance at 4,300 m elevation (USARIEM H02-01, HSRRB A11189). It was conducted by investigators and staff from the Thermal and Mountain Medicine Division of the U.S. Army Research Institute of Environmental Medicine and the Geriatric Research, Education and Clinical Center of the Palo Alto Veterans Administration Health Care System. The project was a multidisciplinary study that allowed several sub-studies to be conducted concurrently without compromising the primary objectives which were to determine: 1) if increased levels of physical activity and antioxidant supplementation will affect the incidence, severity, and duration of Acute Mountain Sickness, markers of oxidative stress and immunity, and variables associated with altitude acclimatization; and 2) if carbohydrate supplementation during prolonged exercise will result in improved performance. Descriptions of the study design, volunteer recruitment, and measurement of energy deficit, exercise protocols, and biochemical studies are described in USARIEM Technical Report No. T-03/07, "Carbohydrate supplementation improves time-trial cycle performance at 4300 m altitude." The sub-study reported here concerns the effects of acute and chronic altitude exposure on pupillary and oculomotor responses and the possible association with Acute Mountain Sickness.

Oculomotor and pupillary reflexes during acute hypoxic exposure were studied on the summit of Pikes Peak, CO (4,300 m) with two different intents. The first objective was to describe the effects of acute hypobaria (subjects breathed 100% oxygen within the first hour of arriving on the summit), followed by changes without supplemental oxygen during the first 24 h of a 14-day exposure. These results and the conclusions of this study phase were reported in USARIEM Technical Report T-03/04, "Oculomotor and pupillary reflexes during acute exposure to hypobaric hypoxia." The second objective, reported here, was accomplished by studying the changes occurring over the next 12 days of the 14-day Pikes Peak exposure with the intent of identifying and characterizing relationships of pupillary and oculomotor reflexes with variables usually associated with altitude acclimatization. The hypotheses tested were that pupillary responses would: 1) be decreased in volunteers receiving antioxidant supplementation; 2) change in concert with urinary catecholamines, and respiratory and circulatory variables; and 3) correlate with the severity of Acute Mountain Sickness.

ACKNOWLEDGMENTS

The authors wish to thank the volunteers for their willingness to endure the difficulties of high altitude exposure and the research procedures they were asked to perform. We also wish to thank the numerous support staff who contributed directly and indirectly to the support of this project: Dr. Beth Beidleman, SGT Tommy J. Bruington, SSG Dan T. Ditzler, LTC Ann Grediagin, Erik Lammi, Sharon K. Leshner, SPC Mona M. Mathow, SPC Jack E. Mazzotti, SGT Dennis M. Rufolo, Tracey J. Smith, Robert Soares, Janet E. Staab, SGT Stephen M. Watt, and Frank Zirpolo.

Special thanks are given to Mr. Mark Sharp and SSG David DeGroot, who ensured that the Pikes Peak Laboratory was made operational and safe and to Dr. Gary Kamimori for his analysis of urinary catecholamines.

EXECUTIVE SUMMARY

Hypoxia caused by travel to high mountain areas can have devastating effects on the health and performance of sensitive individuals. To identify these individuals, an objective index of the hypoxic effect on the central nervous system was studied. In a previous report we described the effects of a 4-h exposure to real and simulated 4,300 m altitude on the pupillary light reflex as measured by the initial pupil diameter (PD), constriction amplitude (CA), and constriction latency (CL). Saccadic velocity (SV) was measured as an index of extraocular motor function. Within 1 h of exposure PD was reduced 5% and CL 2%. After 24 h, the reductions progressed to 12% and 5%, respectively. These effects were readily reversible, not gender related, and independent of eye dominance. The effects were due to the hypoxia *per se* and not the concomitant reduction in barometric pressure. In contrast to pupillary responses, SV, a measure of the rapidity of eye movement, was not acutely affected. We did not test for a relationship with Acute Mountain Sickness (AMS) severity, as peak symptoms occurred well after 4 h of exposure.

To determine whether oculomotor reflexes could serve as an acclimatization variable as well as an objective index of AMS severity, data were collected from the second, long-term phase of the field study conducted on the summit of Pikes Peak. After sea-level baseline (SLB) measurements were taken in Palo Alto, CA, 18 men (25 yrs: range 19-33) were transported to the US Army Maher Memorial Altitude Laboratory, Pikes Peak, CO (4,300 m) where they remained in residence for 14 days. Periodic measurements (days 1-4, 6, 7, 9, 10, and 12) were made of the same pupillary, oculomotor variables (PD, CA, CL, and SV) in addition to classical measures of altitude acclimatization: environmental symptoms questionnaire (ESQ), heart rate (HR), pulse oximetry (SpO_2), end-tidal PO_2 and PCO_2 , and 24-h urinary epinephrine and norepinephrine concentrations.

PD and CL decreased from SLB for first 1-4 days and subsequently returned toward SLB; these changes paralleled changes in ventilatory and circulatory variables. CA decreased on days 1 and 2 and remained decreased for 12 days. SV increased over the first 6 days then returned toward SLB with continued exposure, similar to the changes in urinary catecholamines. With acclimatization, CL correlated with HR and SpO_2 ; SV correlated with PCO_2 , HR, and SpO_2 . AMS severity peaked during days 2-4, returned toward SLB over the next 10 days, and correlated only with CL ($p=0.045$).

Thus, it appears that selected pupillary and oculomotor responses can be used as additional indices of altitude acclimatization, even though there was no strong correlation of any oculomotor measurement with the severity of AMS.

INTRODUCTION

The central nervous system (CNS) is particularly sensitive to diminution of the oxygen levels, nutrient supply, and waste removal. With travel to moderate terrestrial elevations, the primary concern is the ambient partial pressure of oxygen (PO_2) which may be sufficiently reduced so as to initiate a host of physiological and biochemical responses that attempt to compensate for the oxygen reduction, with the end result being the establishment of acclimatization to a specific elevation. However, before complete acclimatization occurs, almost all individuals suffer decrements in physical and mental capacities.

Individuals with varying levels of sensitivity to the reduced ambient oxygen first exhibit the initial signs and symptoms of CNS hypoxia characterized by euphoria, hyperventilation, lightheadedness, giddiness, and extroversion. This is temporally followed by headache, insomnia, nausea/anorexia, fatigue, lassitude, dizziness, insomnia, and malaise (8). The latter are indicative of a complex group of reversible, self-limited symptoms collectively known as Acute Mountain Sickness (AMS). At 4,300 m, AMS symptoms usually do not appear until 6-48 hours after arrival and increase in severity reaching a peak after 48-72 hours, after which they subside over the next 5-7 days. Thus, there appears to be a transition from the initial, primary effect of hypoxia to the secondary, compensatory responses. Both sets of symptoms are indicative of a disturbance in the "internal milieu" of the CNS which may be easily quantified by studying selective components of the visual system.

For many years vision has been recognized as being sensitive to hypoxia. Early quantitative research revolved around advances in aviation and the need to protect pilots and passengers of high-flying aircraft. Various parameters of visual performance have been studied with exposures to hypoxia (10; 10-14; 16; 17). These parameters include dark adaptation, night vision, central brightness contrast, color vision, and central acuity and have a commonality in that they rely to a large extent on volunteer motivation and attention level. Visual tests that are independent of these and other retinal factors are a distinct advantage if they can be shown to relate to subsequent AMS (either as a predictor or a index of severity) and altitude acclimatization. Such tests include oculomotor performance as measured by saccadic velocity and reflexes such as the pupillary response to a light flash. Thus, there is justification for determining whether the measurement of a CNS function that is not dependent on volunteer volition, is affected by hypoxic exposure, and is correlated to the susceptibility and severity of AMS. In addition, it would be beneficial to determine whether changes in oculomotor-hypoxia responses were congruent with the classical measures of altitude acclimatization.

The changes in arterial oxygen saturation, heart rate, PO_2 , and PCO_2 during altitude acclimatization have been well described (21; 22). It has also been recognized for many years that altitude stimulates the sympathetic nervous system (2; 3; 24). All of these variables demonstrate an acute change within minutes or hours, followed by a peak or a nadir after several days, in most cases reaching a different level of equilibrium from that of sea level. These changes are considered the classical physiological indices of altitude acclimatization.

Over the years researchers have attempted to define the reasons for the varied AMS susceptibility and also to identify potential risk factors (1; 7; 18; 19; 23; 25; 27). But, to date, no one has been able to predict, with reasonable certainty, who will become ill and to what extent. It has also been difficult to identify objective physiological variables that may be associated with illness. Without these relevant objective variables, research studies seeking to evaluate effective pharmaceutical or strategic beneficial interventions are decidedly more difficult. To be able to predict and quantify AMS in a more expeditious manner would be a distinct advantage, as it may eventually be possible to configure a military unit that would be able to function at altitude with minimal incapacitation by altitude illness. Achieving success in this goal would be accomplished by identifying simple, rapid, and objective physiological variables that are responsive to hypoxia – ones that follow the same temporal and quantitative pattern as reported AMS symptoms.

The purpose of this investigation was to extend the study of the effects of exposures to high altitude (4300 m, ~459 mmHg) on oculomotor performance in an effort to determine a correlation with: 1. the classic measures of altitude acclimatization, and 2. the severity of AMS.

METHODS

STUDY DESIGN AND PIKES PEAK ASCENT

As part of research protocol (H02-01) investigating the effect of increase energy expenditure, antioxidant supplementation, and carbohydrate ingestion on work performance and altitude acclimatization to 4300m altitude, 18 healthy male volunteers¹ were recruited from the vicinity of Palo Alto, CA during Jan-April 2002. Requisite inclusion criteria were: 18-35 years old, non-smokers, normal height-to-weight ratios (BMI = 20-27), no large weight changes within the last 6 months, born below 5000 ft (~1,530 m, Denver, CO), residence at or near sea level for 6 months prior to the study, no ingestion of non-approved vitamins and food supplements, and ability to perform cycle exercise for at least 1 hour at ~ 75% of their age-predicted maximal heart rate. Antioxidant supplementation consisted of a mixture of β -carotene, α -tocopherol, ascorbic acid, selenium, and zinc administered 3 weeks prior to ascent and during the 12 exposure days.

Sea-level, baseline measurements were made at the Palo Alto Veterans Affairs Health Care System, Palo Alto, CA during May and June 2002. Diet, caloric expenditure, and exercise studies are described in USARIEM Technical Report T-03/07 (5). Every 1-3 days during July and August, one or two subjects were transported to Colorado Springs, CO where they spent the afternoon, evening, night, and next morning breathing supplemental oxygen (96%) supplied by an oxygen concentrator (AirSep, Buffalo, NY). During their automobile ascent to the summit of Pikes Peak (1.5-hour) subjects breathed 100% bottled oxygen. After reaching the summit and until they

¹ Only men were included in the main study because the antioxidant properties of estrogen could confound one of the major objectives of the study which was to determine the effect of antioxidant supplementation on oxidative stress markers during exercise at altitude.

completed 3 consecutive pupillometry/oculomotor tests (~1 hour), they continued to breathe oxygen from another concentrator. Volunteers repeated the pupillometry/oculomotor tests 3 h after removal of the supplemental oxygen. These results, which represent the effects of hypobaric normoxia and acute hypobaric hypoxia, are reported in USARIEM Technical Report T-03-04 (4).

A total of 18 men (25 yrs: range 19-33) were transported to the US Army Maher Memorial Altitude Laboratory, Pikes Peak, CO (4,300 m). Periodic measurements (days 1-4, 6, 7, 9, 10, and 12) were made of oculomotor variables (PD, CA, CL, and SV) and the classical measures of altitude acclimatization: heart rate (HR), pulse oximetry (SaO_2), end-tidal PO_2 and PCO_2 , and 24-h urinary epinephrine and norepinephrine concentrations. Urinary catecholamines were measured up until day 10.

Conditions at the summit within the USARIEM Maher Memorial Laboratory averaged 460 ± 2 mmHg, 20-22°C and 15-20% relative humidity. Ambient illumination level at Palo Alto and at the summit laboratory was approximately 230-245 lux during the testing day.

INSTRUMENTATION

Oculomotor and pupillary reflexes were made using the FIT 2000 screener (PMI, Inc., Rockville, MD). The Fit 2000 is a self-contained, fully automated Class I medical instrument designed to analyze the eye reflexes for signs of neurological changes due to a variety of positive or negative stimuli. The instrument can detect changes as small as 0.05 mm in pupil size and eye movements as small as one degree. Subjects determined left or right eye dominance prior to any testing and always used the dominant eye for all measurements. Initial pupil diameter was determined prior to any light flashes. To evoke a pupil reflex, 4-7 controlled, 1-s flashes of constant intensity light were produced allowing measurement of constriction latency and amplitude. To evoke a measured saccadic eye movement, a lighted target moved along a precise horizontal path that the volunteer followed with his/her dominant eye. The entire sequence of 4 measurements took ~30 s to complete. The mean of three sequences, taken 2-4 min apart, was used as a data point.

Blood oxygen saturation (SaO_2) and heart rate (HR) were measured by pulse oximetry (Nellcor N-200, Nellcor Inc., Pleasanton, CA) after the volunteers were seated and rested for 10 min. Resting oxygen uptakes and carbon dioxide production were measured using a metabolic cart (True Max 2400, ParvoMedics, Salt Lake City, UT).

Urinary catecholamines, norepinephrine and epinephrine, were measured from 24-h urine collections. Catecholamines were isolated by alumina extraction using a Chromsystems reagent kit (Alko Diagnostics, Holliston, MA). Once extracted, catecholamine concentration was quantified by high performance liquid chromatography HPLC system (Waters Corp., Milford, MA). Data were stored and analyzed using the Waters Millennium software package (V 2.10). The sensitivity of the assay was 5 pg on column with a signal to noise ratio of 5 to 1, a between days coefficient of variation of less than 2%, and a within days variation of less than 1%. The standard curve for the range of 5-5000 pg had a correlation coefficient of 0.998.

The severity of Acute Mountain Sickness (AMS) was assessed using a weighted average score of selected cerebral symptom determined from the Environmental Symptoms Questionnaire (ESQ). The ESQ is a validated, self-reported, 67-question symptom inventory used to determine the presence and severity of symptoms due to exposures to heat, cold, and hypoxia (26). A weighted factor score of 11 subjective symptom scores was used; a value of 0.7 or greater is indicative of the presence of AMS.

STATISTICS

Test-retest reliability analyses were performed for the 4 oculomotor and pupillary parameters (initial pupil diameter, constriction amplitude, constriction latency, and saccadic velocity) using the FIT 2000. Simple regression analysis was used on previously collected data from 37 subjects from a previous study, H98-09, "Effect of residence at low and moderate altitudes on arterial oxygen saturation at moderate-to-high altitudes." Subjects were tested at the beginning and end of a 1-h rest period under sea-level conditions. The degree in which data values are consistent over time is an indication of the variation from testing session to testing session, i.e., variable variability. Coefficient of stability (r) is the measure of test-retest reliability.

Initial analyses utilized a 2x10 repeated measures of analysis of variance (group x time): (antioxidant and placebo x SL, D1, D1.25, D2, D3, D4, D6, D7, D9, D10, D12) with 9 subjects in each group (Sigma Stat, v2.03, SPSS, Chicago, IL). If no significant effect due to antioxidant was found, both groups were combined and treated as one.

When significant main effects were found ($P \leq 0.05$), post-hoc analyses using Dunnett's (for multiple comparisons versus control or Tukey's (for all pairwise comparisons) tests were utilized. Regression analyses of oculomotor performance and pupillary reflexes vs. acclimatization variables (catecholamine excretion, PO_2 and PCO_2 , heart rate, blood oxygen saturation) were performed using a linear, repeated measures regression test with between subject differences taken into account with dummy variables used to encode different subjects (6).

RESULTS

COLLAPSING GROUPS

There were no significant differences found in any of the four measured oculomotor variables (initial pupil diameter, constriction amplitude, constriction latency, and saccadic velocity) regardless of the group (antioxidant or placebo) into which the subjects were placed; therefore, all the subjects were combined into one group.

RELIABILITY

Test-retest reliability for the initial pupil diameter measurement is shown In Figure 1 and serves as an example of the variability of all the oculomotor variables. Values for the remaining oculomotor and pupillary reflex variables are shown in Table 1. Reliability values for initial pupil diameter, constriction amplitude, and constriction latency are all

similar (0.92 to 0.80). Saccadic velocity showed the worst reliability with an r value of 0.54.

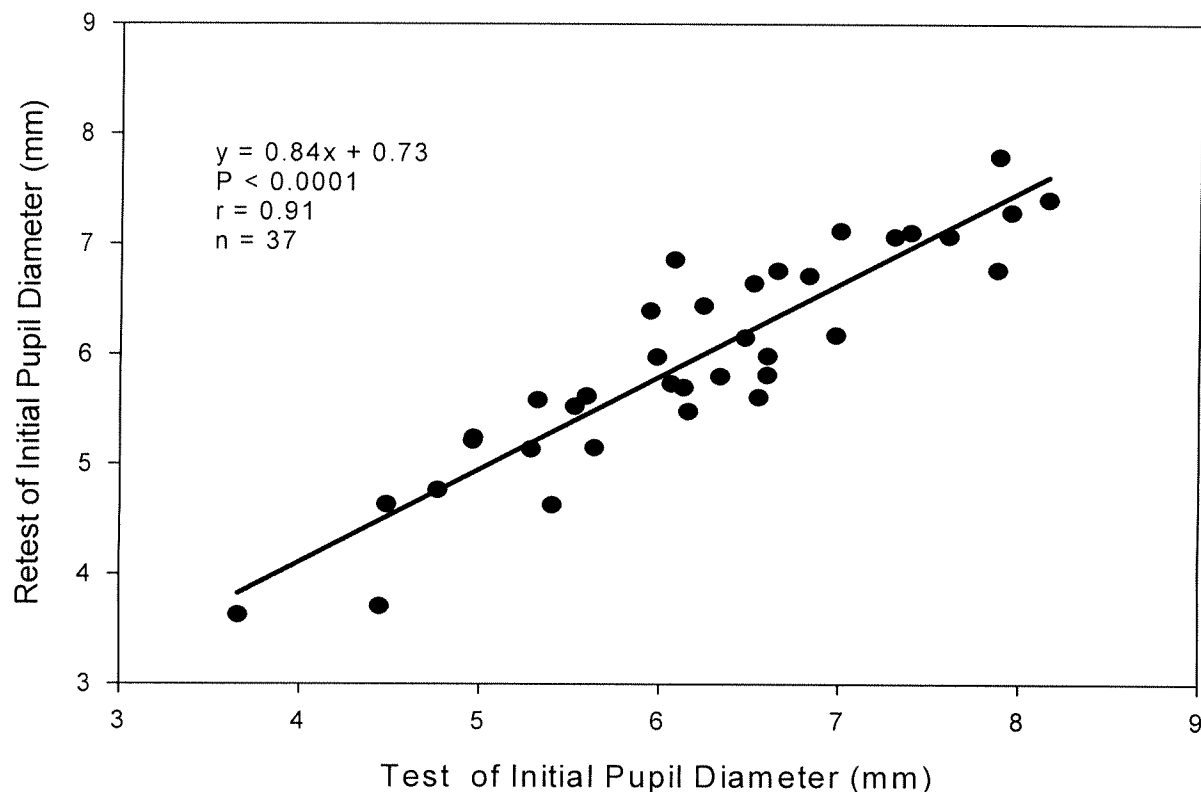


Figure 1. Test-retest reliability of initial pupil diameter as an example of pupillary reflex variables. Each data point represents the mean of a set of three initial pupil diameter (PD) measurements from one subject taken one hour apart.

Table 1. Test-retest characteristics of 4 oculomotor pupillary reflex variables

Variable	N	Slope	Intercept	P	R
PD (mm)	37	0.84	0.73	<0.0001	0.91
CA (mm)	37	0.91	20.3	<0.0001	0.92
CL (msec)	37	0.80	0.23	<0.0001	0.80
SV (mm/msec)	37	0.46	36.1	=0.0006	0.54

N = number of subjects; P= probability value; r = reliability coefficient; PD = initial pupil diameter; CA = constriction amplitude; CL = constriction latency; SV = saccadic velocity

OCULOMOTOR RESPONSES DURING ACCLIMATIZATION

The changes in initial pupil diameter and pupil constriction amplitude during 12 days of continuous exposure to 4300 m are shown in Figure 2. Pupil diameter (A) decreased during the first 3 days followed by a return to the initial sea-level baseline values. Constriction amplitude (B) also decreased during the first 2 days but appeared to establish a new, albeit lower stable level evident after 9 and 12 days.

Constriction latency (Figure 3A) showed a similar response as initial pupil diameter, decreasing during the first 3 days of exposure followed by a return toward sea-level values. Saccadic velocity (3B), never decreased below sea-level values and, in fact, remained elevated or slightly elevated for the entire exposure.

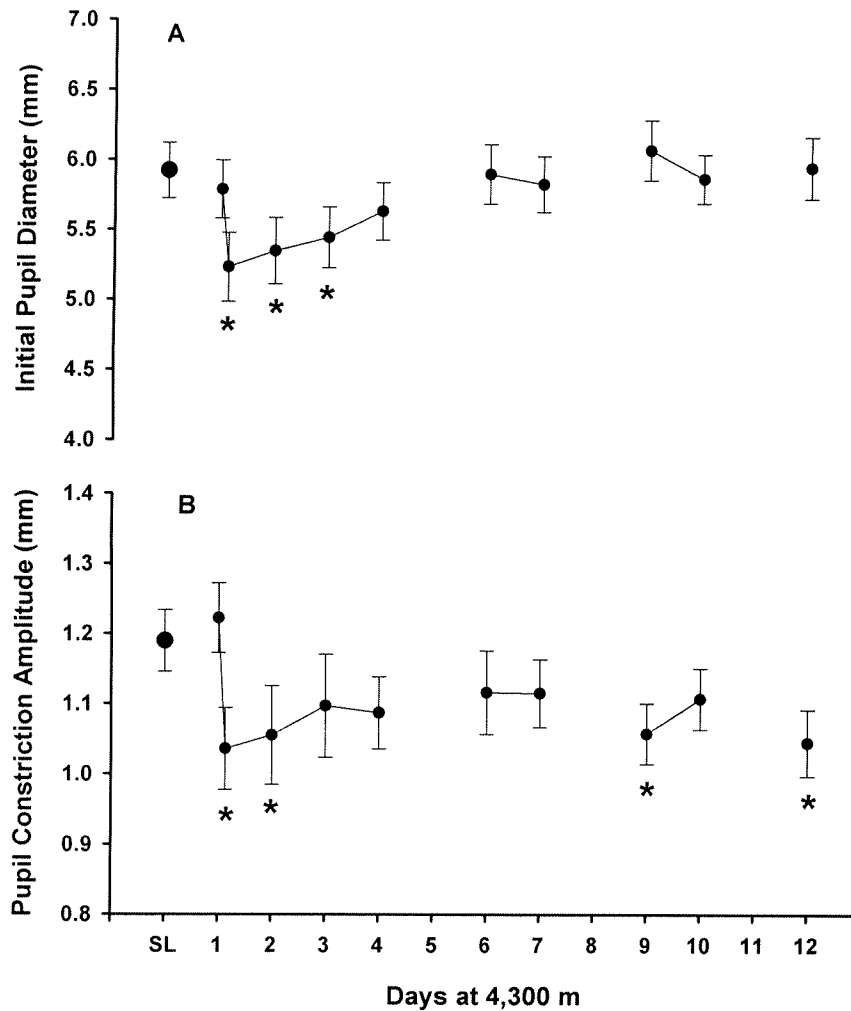


Figure 2. Initial pupil diameters (A) and constriction amplitudes (B) measured at sea level, immediately upon exposure to 4,300 m altitude, and periodically during acclimatization. Values are mean \pm S.E. * = significantly different from sea level ($p \leq 0.05$), $n=17$.

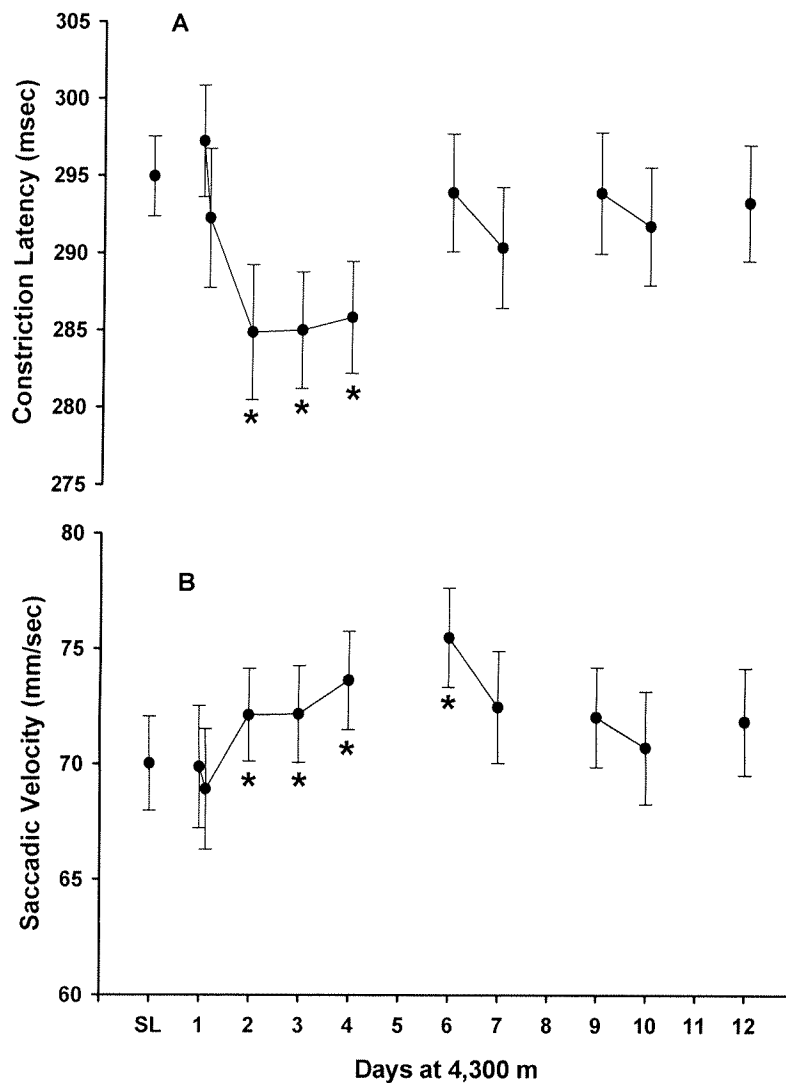


Figure 3. Constriction latencies (A) and saccadic velocities (B) measured at sea level, immediately upon exposure to 4,300 m altitude, and periodically during acclimatization. Values are means \pm S.E. * = significantly different from sea level, $p \leq 0.05$, $n=17$.

CARDIOVASCULAR AND NEUROENDOCRINE RESPONSES AND ACUTE MOUNTAIN SICKNESS

Figures 4-6 illustrate classic changes in PO_2 , PCO_2 , heart rate, arterial oxygen saturation, and daily catecholamine excretion levels that occur during continuous exposure to 4,300 m altitude. Respiratory responses show an acute change over the first 4 days followed by the establishment of new plateaus. During 12 of the 14 days of exposure values never approach their initial sea-level value. Increases in resting heart rate peaked during days 2-4, followed by a slow decrease toward sea-level baseline values; arterial oxygen saturation showed similar but reciprocal changes with saturation levels stabilizing at approximately 89% after 10-12 days of exposure. Urinary catecholamine concentrations mirrored changes in resting heart rates, except that

concentrations after 5-7 days, although elevated, were not significantly different from sea level.

The progression and improvement of AMS symptom severity is depicted in Figure 7. Symptom severity rose on day 1, peaked during days 2-4, and gradually returned toward sea level thereafter.

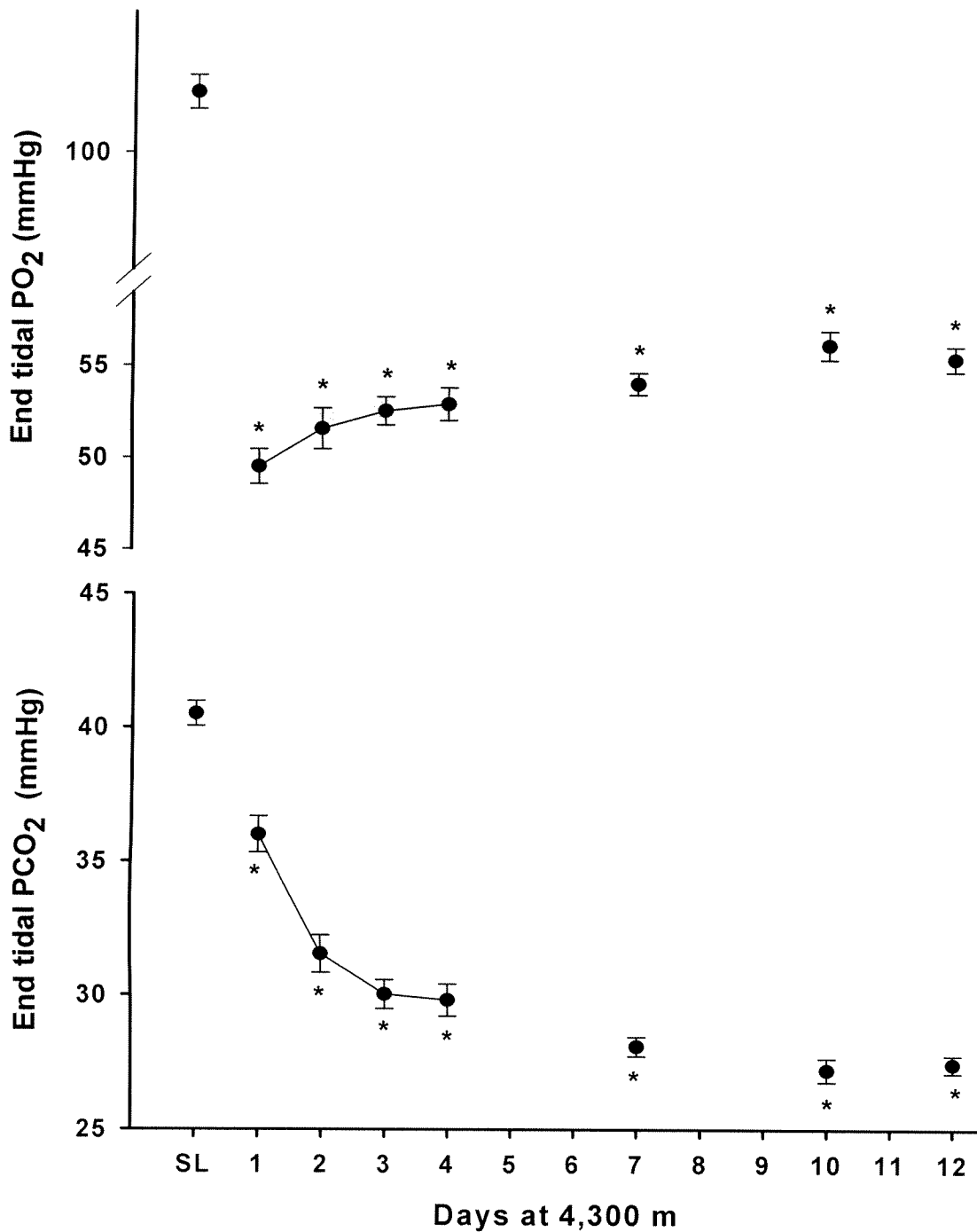


Figure 4. End-tidal partial pressures of O₂ and CO₂ at sea level (SL) and during acclimatization to 4,300 m altitude. Values are means \pm S.E.; n = 17; * = significantly different from sea level, p \leq 0.05.

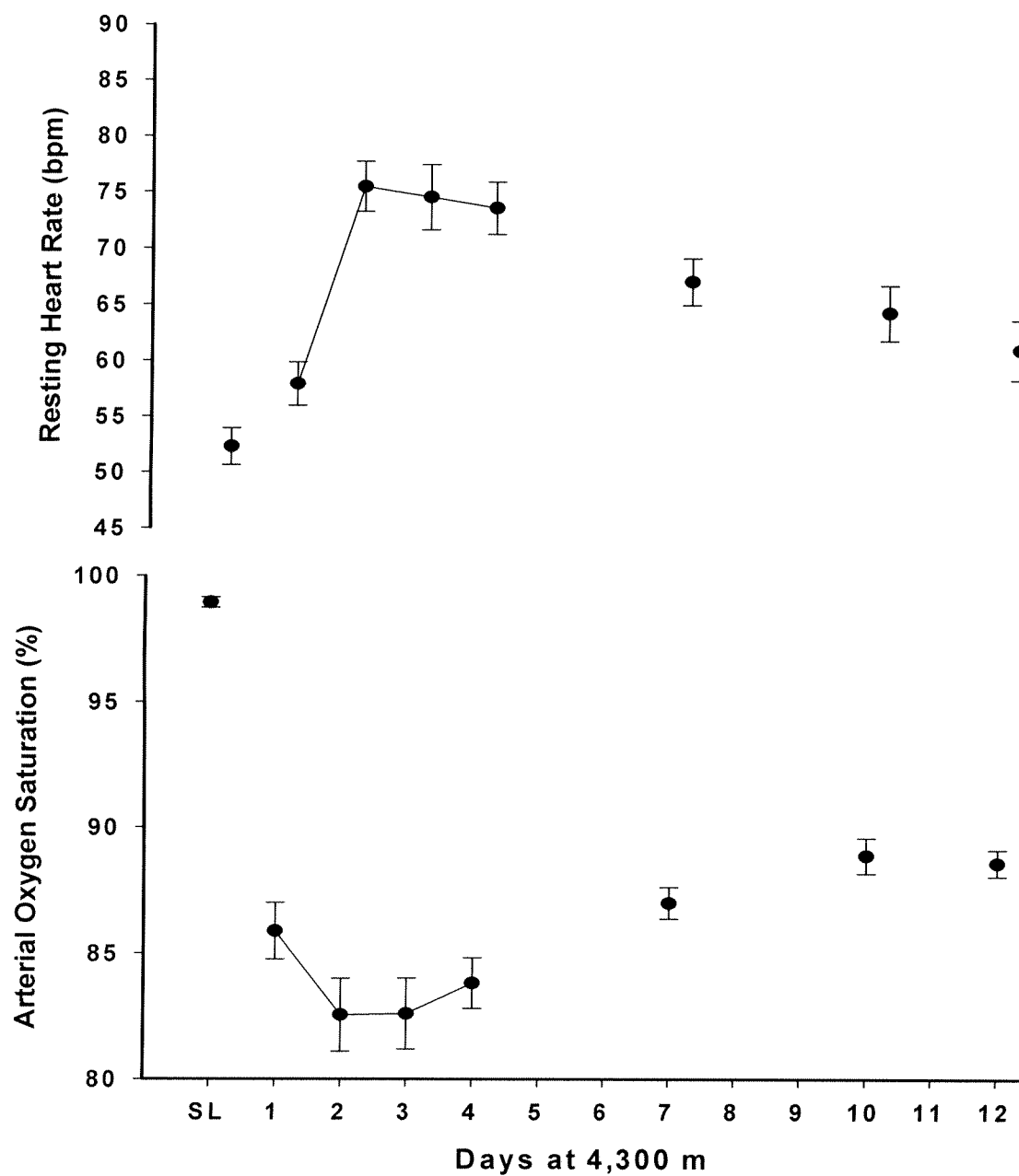


Figure 5. Changes in resting heart rate and arterial oxygen saturation during 12 days of exposure to 4,300 m. Values are means \pm S.E. * = significantly different from sea level, $p \leq 0.05$, $n=17$.

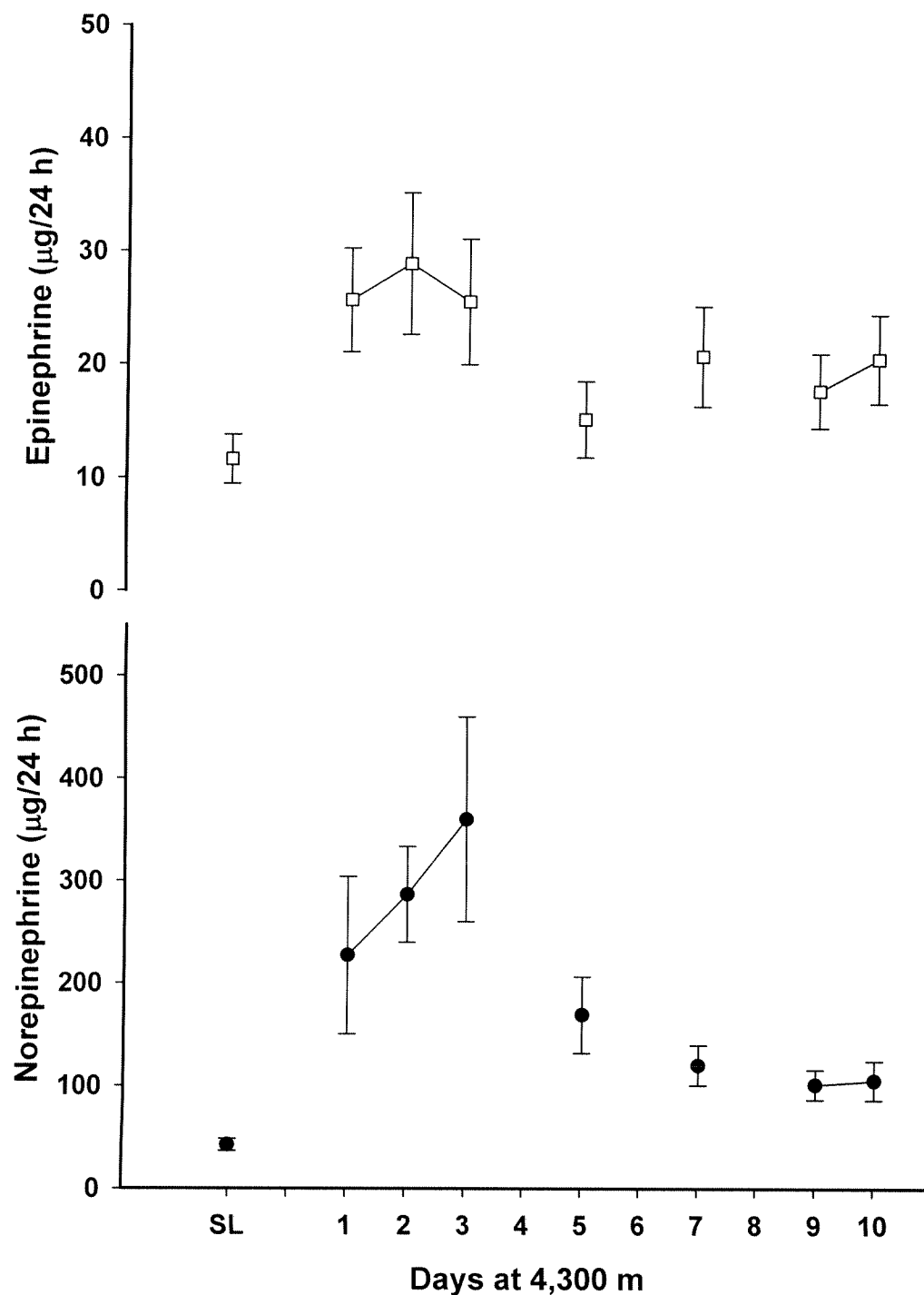


Figure 6. Twenty-four hour urinary catecholamine excretion rates at sea level (SL) and during acclimatization to 4,300 m altitude. Values are means \pm S.E.; $n = 17$; * = significantly different from sea level, $p \leq 0.05$.

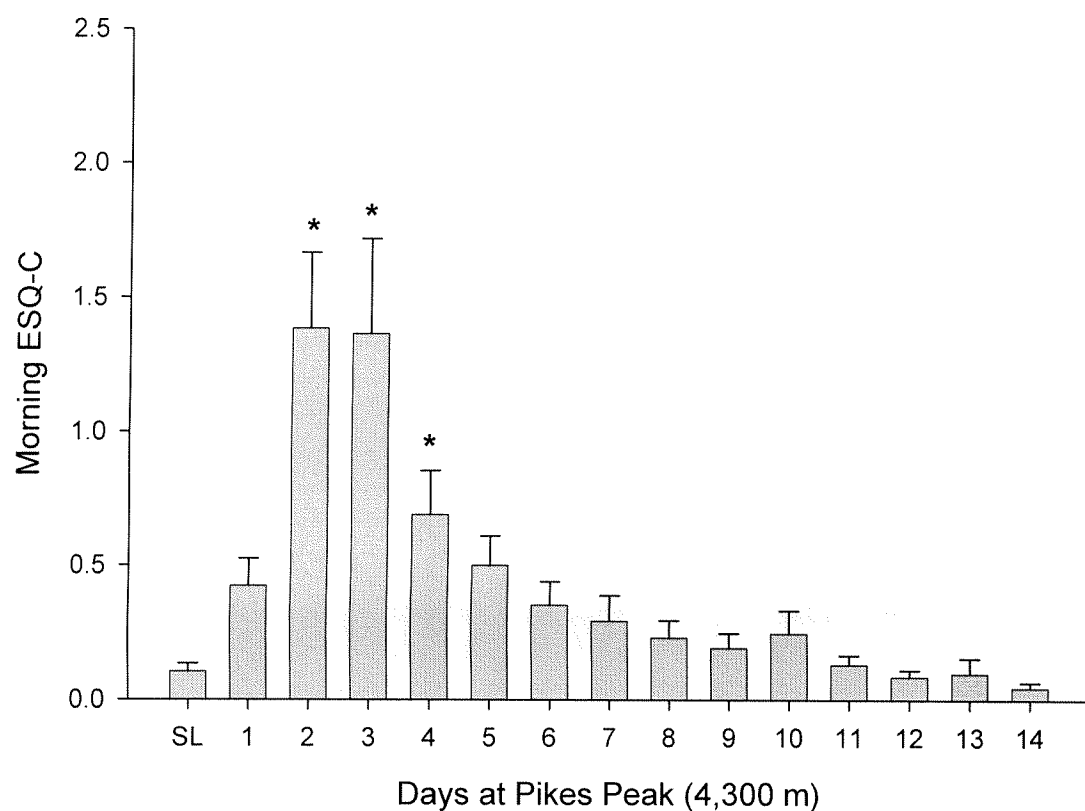


Figure 7. Acute Mountain Sickness (AMS) cerebral symptoms derived from morning environmental symptoms questionnaires (ESQ-C) during acclimatization to 4,300 m altitude. * $P < 0.001$, means \pm SE, $n = 18$ (< 6 days); $n = 17$ (> 5 days). Day 1 = ~1h after removal of supplemental O_2 which was administered during ascent and initially on the Pikes Peak summit: Day 2 and thereafter are progressive 24-h increments.

CORRELATION OF OCULOMETRIC VARIABLES AND PHYSIOLOGICAL INDICES OF ACCLIMATIZATION

Table 2. Summary of regression coefficients comparing oculomotor variables with classic parameters of acclimatization during the 1st 3 days of exposure to 4,300 m altitude.

Acclimatization Variable	Initial Pupil Diameter (mm)		Constriction Amplitude (mm)		Constriction Latency (msec)		Saccadic Velocity (mm/sec)	
	Slope±SE	P	Slope±SE	P	Slope±SE	P	Slope±SE	P
NE (μg/24 h)	0.00±0.00	ns	0.00±0.00	ns	-0.01±0.01	ns	0.00±0.00	ns
EPI (μg/24 h)	0.01±0.01	ns	0.00±0.02	ns	0.07±0.14	ns	0.01±0.07	ns
PO ₂ (mmHg)	0.01±0.03	ns	0.00±0.01	ns	0.71±0.78	ns	0.80±0.34	0.028
PCO ₂ (mmHg)	0.01±0.02	ns	0.00±0.01	ns	0.91±0.53	0.099	-0.81±0.21	0.001
Heart rate (bpm)	0.00±0.01	ns	0.00±0.00	ns	-0.47±0.14	0.003	0.21±0.07	0.006
SpO ₂ (%)	0.00±0.02	ns	0.00±0.01	ns	1.52±0.27	0.001	-0.32±0.18	0.009

NE = urinary norepinephrine; EPI = urinary epinephrine; PO₂ = end-tidal partial pressure of oxygen; PCO₂ = end-tidal partial pressure of carbon dioxide; SpO₂ = pulse arterial oxygen saturation; ns = not significant.

In order to determine whether the changes observed in oculometrics coincided with changes in the variables classically used to monitor altitude acclimatization, a repeated measured regression method was used so that all the data collected during acclimatization could be used in the analysis. During altitude acclimatization constriction, latency coincided with changes in heart rate and SpO₂, while saccadic velocity coincided with the latter two variables and also PO₂ and PCO₂ (Table 2).

ASSOCIATION OF OCULOMETRICS AND AMS

Repeated measures regression of oculometrics from data obtained on days 2-4 (when AMS was at its severest in most subjects) indicated very little association of oculometrics with the severity of AMS. AMS severity was associated with an increase in constriction latency ($p=0.045$) and possibly a decrease in saccadic velocity ($p=0.066$).

When linear regression analysis is performed using AMS severity from day 3 and the four oculometric variables no correlation was found, even when analyzed as a difference from sea-level baseline or as a % change from baseline on day 3.

DISCUSSION

The objective of the present report was to determine the effect of altitude acclimatization on oculomotor performance and pupillary reflexes. In order to accomplish this objective, it was necessary to verify that repeated measurements of the four oculomotor variables would be consistent and reliable when performed repeatedly in the same individual. Our results indicated that measurements of pupillary reflexes and oculomotor performance were consistent and reliable. We also determined that there was no significant effect between the antioxidant and the placebo groups, allowing us to combine data from all the subjects into one group, thus increasing the statistical power to determine any relationship between AMS and oculometrics.

Confirming the results from our previous report (4), we observed that initial pupil diameters, constriction amplitudes, and constriction latencies were reduced with initial hypoxic exposures (>1 h), while saccadic velocities were increased during the same time period. We extended the time course by observing that the reductions persisted for the first 2-4 days and then returned toward baseline levels, except for constriction amplitude which tended to remain reduced for the entire 12 days. Saccadic velocities increased during the first 6 days but then returned toward baseline. These observations tended to follow the changes seen in PO_2 , PCO_2 , heart rate, saturation, and catecholamines, i.e. an acute change during the first 2-4 days followed by the establishment of a new stable baseline or a return to levels indistinguishable from baseline. The development and remission of AMS symptoms followed its classical pattern – an increase the first day, peaking during days 2-4, followed by a rapid return to baseline levels.

The question of correspondence of oculometric variables with physiological acclimatization variables may be difficult to assess because no previous studies have attempted to relate oculometrics and altitude acclimatization. There are two reports that describe pupillary changes during acute hypoxia. Van der Post (29) found decreases in saccadic velocity in 12 volunteers exposed to 130 min of a hypoxic gas mixture producing arterial O_2 saturations of ~80%. In patients with different degrees of respiratory insufficiency or during anesthesia, Jordanov and Ruben (9) observed either no change or pupillary contraction within minutes of exposure to arterial PO_2 's as low as 34 mmHg. Our results indicated that all the measured oculometric variables were affected by hypoxia within the first 3-4 days of altitude exposure. Initial pupil diameters and constriction latencies were depressed in the first 24 h (day 2) of exposure, but returned toward normal thereafter. Constriction amplitude remained decreased for almost the entire 12 days. Saccadic velocities were increased from days 2 to 6 when oxygen saturation was 83% but returned to levels indistinguishable from baseline measurements. All these oculometric changes temporally corresponded to the changes in heart rate, PO_2 , PCO_2 , and pulse oxygen saturation which also showed the greatest changes during the first 4 exposure days. Significant correlative changes were observed between constriction latency and heart rate and SpO_2 . Saccadic velocity also correlated with changes in PCO_2 , heart rate, and pulse oxygen saturation during acclimatization. Thus, it appears that selected pupillary and oculomotor measures can

be used as additional indices of altitude acclimatization, even though there was no strong correlation of any oculomotor measurement with the severity of AMS.

A physiological explanation for these oculometric changes with hypoxic exposure and acclimatization is difficult to provide. There appears to be two phenomena occurring simultaneously: reductions in pupillary diameters and constriction amplitudes indicative of a central depressive effect of hypoxia; and reductions in constriction latencies (faster reaction times) and increased saccadic velocities both of which are indicative of a stimulatory effect. Approaches that can be taken to explain the dichotomy are: a direct, transient, and inhibitory effect of hypoxia on specific components of the central nervous system; and an indirect effect due to increases in peripheral neurohumoral factors; or a combination of both occurring at the same time.

A healthy iris is in constant motion even when light conditions and accommodation are constant. This physiologic pupillary instability or dynamic equilibrium is due to fluctuations in the activity of sympathetic and parasympathetic innervations to the iris muscles. As explained by Thompson (28), when the level of consciousness or alertness is reduced, as may also occur with hypoxia, pupils become smaller and oscillation becomes apparent. This is a reflection of a decrease in inhibition of the Edinger-Westphal nucleus, a parasympathetic component of the oculomotor nucleus, followed by a preponderance of parasympathetic outflow. There is further evidence for a reduction in pupil size with a decrease in level consciousness, alertness, and acute hypoxia. Morad et al. (20) observed decreases in pupil size with fatigue and subjective feelings of sleepiness. Lowenstein et al. (15) observed that miosis (pupil contraction) is common and light reflexes are markedly reduced when subjects are tired. Thus, the pupillary hypoxic response (miosis) may be analogous to the pupillary response preceding sleep or during fatigue and could result from a decrease in inhibition of a CNS parasympathetic nucleus.

The decrease in the amplitude of the light reflex is coupled to the decrease in pupil diameter in that light reflexes are superimposed on the dynamic equilibrium. This superimposition can be modified by humoral adrenergic factors (i.e., peripheral catecholamines) as well as mechanical limitations of the iris. Peripheral changes in catecholamine levels probably have little effect on pupillary reflexes because their concentrations result primarily from the accumulation of unmetabolized catecholamines and spillover from neuronal terminals in sympathetic tissues and the adrenal medulla. Thus, plasma and urine levels provide a limited, delayed view of the possible systemic effects at peripheral nerve endings and in the CNS. If pupil diameter is sufficiently reduced, it is conceivable that the amplitude of a light response would also be reduced if the ability of the pupil to constrict further is limited. This mechanical contraction limitation is not likely. The mean pupil diameter at sea level in our subjects was 5.92 mm and the mean minimum diameter on the first day of altitude exposure was 5.23 mm. Thus, with a normal range of 1 to 9 mm we do not believe that a mechanical limitation was present. Nevertheless, the same flash stimulus could elicit a smaller response if a nonlinear length-tension relationship is involved. Therefore, at the present time, we cannot explain the decrease in initial pupil diameter and decrease in flash-produced constriction amplitude that occurs during altitude acclimatization.

It is possible that peripheral catecholamines could affect specific pupillometric measurements. The changes observed in constriction latencies and saccadic velocities are indicative of a stimulation of the sympathetic nervous system, i.e., reaction times were faster (shorter constriction latencies) and increased saccadic velocities. Saccadic velocity, as measured by the FIT 2000, is the outcome movement of a horizontal, voluntary reflex arc involving simultaneous contraction and relaxation of agonist and antagonist muscles (medial and lateral recti muscles). These muscles, being extraocular and striated, may be more easily affected by blood-borne catecholamines. Both constriction latency and saccadic velocity were significantly correlated with heart rate and pulse oxygen saturation; saccadic velocity was also correlated with end-tidal PCO_2 . However, there was no correlation with urinary catecholamines.

Although the progression of AMS severity showed classic quantitative and temporal changes with altitude acclimatization at 4300 m (8), we were unable to demonstrate any strong correlation with the measured oculometrics. In a previous study, we were able to show that the decrease in pupil diameter was correlated with the decrease in arterial oxygen saturation (4) and low arterial oxygen saturation has been correlated with subsequent severity of AMS (25). Nevertheless, we were unable to correlate changes in pupil diameters to changes in AMS even though the majority of subjects (10 of 18 on altitude day 3) indicated they had symptoms severe enough to be considered AMS. In fact, we were only able to demonstrate a significant positive correlation of AMS severity with constriction latency and a possible negative correlation with saccadic velocity.

CONCLUSIONS

A host of respiratory, cardiovascular, endocrine, and neuroendocrine changes are invoked by ascent to high terrestrial altitudes. Ocular and pupillary responses also appear to be affected by hypoxia with changes characterized by an acute response over several days followed by stabilization to a previous baseline or the establishment of a new baseline. The first 4-6 days of exposure are characterized by decreases in initial pupil diameter, constriction amplitude, and constriction latency. In contrast, saccadic velocity, an extraocular striated muscle function, increases during the same time period. The latter two variables may represent an indirect hypoxic effect due to increased sympathetic activity while the former two variables may be associated with a direct effect of hypoxia on the CNS. However, no correlation could be consistently demonstrated with the severity of Acute Mountain Sickness. Thus, it appears that selected pupillary and oculomotor measures can be used as additional indices of altitude acclimatization, even though there was no strong correlation of any oculomotor measurement with the severity of AMS.

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